```
=> d his  l1-l18

     (FILE 'CAPREVIEWS' ENTERED AT 07:58:00 ON 10 JUL 95)
               DEL HIS Y

     FILE 'HCA' ENTERED AT 08:03:17 ON 10 JUL 95
               E KOHN D/AU
L1          35 S E3-6
               E KOHN DONALD/AU
L2          21 S E3-4
               E BLAESE, M/AU
               E BLAESE M/AU
L3           9 S E3-5
               E MULLEN C/AU
L4          14 S E3-6 OR E11
               E MOEN R/AU
L5          25 S E4-6 OR E9-11
L6         100 S L1 OR L2 OR L3 OR L4 OR L5
               SAVE L6 TEMP MILNE/A
L7         854 S (( RETRO (L) VIR?) OR RETROVIR?)(L) VECTOR#
L8        2267 S ADENOSINE DEAMINASE#
L9          48 S IMMUNE DEFICIENCY (L) COMBIN?
L10         21 S SEVERE COMBINED IMMUNE DEFICIENCY
L11        245 S CD34
L12         18 S L6 AND (L7 OR L9 OR L10 OR L11)
L13          8 S L6 AND L8
L14         21 S L13 OR L12      inventor search
L15       5454 S RETROVIR? OR RETRO (L) VIR?
L16          2 S L15 AND L8 AND L11
L17         75 S L15 AND L8
L18         15 S L15 AND L11

=> fil hca
FILE 'HCA' ENTERED AT 08:48:45 ON 10 JUL 95
USE IS SUBJECT TO THE TERMS OF YOUR CUSTOMER AGREEMENT
COPYRIGHT (C) 1995 AMERICAN CHEMICAL SOCIETY (ACS)

FILE COVERS 1967 - 8 Jul 1995 (950708/ED)  VOL 123 ISS 2

HCAPLUS IS NOW ONLINE!
'OBI' IS DEFAULT SEARCH FIELD FOR 'HCA' FILE

=> d bib ab l14 1-21;d bib abs hitind l16 1-2

L14  ANSWER 1 OF 21  HCA  COPYRIGHT 1995 ACS
AN   123:7845  HCA
TI   Retrovirally marked CD34-enriched peripheral blood and
     bone marrow cells contribute to long-term engraftment after
     autologous transplantation
AU   Dunbar, Cynthia; Cottler-Fox, Michelle; O'Shaughnessy, Joyce A.;
     Doren, Sandra; Carter, Charles; Brenson, Ronald; Brown, Sherri;
     Moen, Robert C.; Greenblatt, Jay; et al.
CS   Hematol. Branch, Natl. Institutes Health, Bethesda, MD, USA
SO   Blood (1995), 85(11), 3048-57
     CODEN: BLOOAW; ISSN: 0006-4971
```

DT Journal
LA English
AB A preliminary human autologous transplantation study of retroviral gene transfer to bone marrow (BM) and peripheral blood (PB)-derived CD34-enriched cells is described. Eleven patients with multiple myeloma or breast cancer had cyclophosphamide and filgrastim-mobilized PB cells CD34-enriched and transduced with a retroviral marking vector contg. the neomycin resistance gene, and CD34-enriched BM cells transduced with a second marking vector also contg. a neomycin resistance gene. After high-dose conditioning therapy, both transduced cell populations were reinfused and patients were followed over time for the presence of the marker gene and any adverse effects related to the gene-transfer procedure. All 10 evaluable patients had the marker gene detected at the time of engraftment, and 3 of 9 patients had persistence of the marker gene for greater than 18 mo post-transplantation. The marker gene was detected in multiple lineages, including granulocytes, T cells, and B cells. The source of the marking was both the transduced PB graft and the BM graft, with a suggestion of better long-term marking originating from the PB graft. The steady-state levels of marking were low, with only 1:1000 to 1:10,000 cells pos. There was no toxicity noted, and patients did not develop detectable replication-competent helper virus at any time post-transplantation. These results suggest that mobilized PB cells may be preferable to BM for gene therapy applications and that progeny of mobilized peripheral blood cells can contribute long-term to engraftment of multiple lineages.

L14 ANSWER 2 OF 21 HCA COPYRIGHT 1995 ACS
AN 122:129910 HCA
TI Multiple modifications in cis elements of the long terminal repeat of **retroviral vectors** lead to increased expression and decreased DNA methylation in embryonic carcinoma cells
AU Challita, Pia-Maria; Skelton, Dianne; El-Khoueiry, Anthony; Yu, Xiao-Jin; Weinberg, Kenneth; **Kohn, Donald B.**
CS Dep. Microbiol. Pediatrics, Univ. Southern California Sch. Med., Los Angeles, CA, USA
SO J. Virol. (1995), 69(2), 748-55
CODEN: JOVIAM; ISSN: 0022-538X
DT Journal
LA English
AB Infection by murine retroviruses in embryonic carcinoma (EC) and embryonic stem cells is highly restricted. The transcriptional unit of the Moloney murine leukemic virus (MoMuLV) long terminal repeat (LTR) is inactive in EC and embryonic stem cells in assocn. with increased proviral methylation. In this study, expression in F9 EC cells was achieved from novel retroviral vectors contg. three modifications in the MoMuLV-based retroviral vector: presence of the myeloproliferative sarcoma virus LTR, substitution of the primer binding site, and either deletion of a neg. control region at the 5' end of the LTR or insertion of a demethylating sequence. The authors conclude that inhibition of expression from the MoMuLV LTR in EC cells is mediated through the additive effects of multiple cis-acting elements affecting the state of methylation of the

provirus.

L14 ANSWER 3 OF 21 HCA COPYRIGHT 1995 ACS
AN 122:98089 HCA
TI **Retroviral vectors** containing chimeric promoter/enhancer elements exhibit cell-type-specific gene expression
AU Couture, Larry A.; **Mullen, Craig A.**; Morgan, Richard A.
CS National Heart, Lung, and Blood Institute, Bethesda, MD, 20892, USA
SO Hum. Gene Ther. (1994), 5(6), 667-77
CODEN: HGTHE3; ISSN: 1043-0342
DT Journal
LA English
AB Retroviral vectors were constructed in which the U3 promoter/enhancer of Moloney murine leukemia (MoMLV) was replaced by the corresponding region from five related murine retroviruses - AKR murine leukemia virus (AKV), Harvey murine sarcoma virus (HaMSV), myeloproliferative sarcoma virus (MPSV), SL3-3, and the NZB-xenotropic virus (Xeno). In these vectors the chimeric long terminal repeat (chLTR) drives the expression of the chloramphenicol acetyl transferase (CAT) reporter gene that is followed by an internal SV40 virus early region promoter linked to the neomycin phosphotransferase II (NEO) gene. As an initial measure of the relative promoter/enhancer strength of the chLTR vectors, the murine NIH-3T3 cell line and the human JURKAT cell lines were transfected and assayed for CAT reporter activity. Relative to the MoMLV vector, the HaMSV construct was the most active in NIH-3T3 cells whereas the SL3-3 vector displayed the greatest activity in JURKAT cells. Retroviral vector producer cell populations and cell clones were established for each chLTR vector, and all were capable of yielding high vector titers (>105 G418R cfu/mL on NIH-3T3). Supernatant from these cells was used to transduce both mouse and human cell lines and primary cells. In NIH-3T3 cells and two murine fibrosarcoma cell lines, the HaMSV chLTR vector was slightly more active than the MoMLV chLTR vector. In the human HepG2 and HeLa cell lines, the MPSV chLTR vector was the most active. Data from the human JURKAT T-cell line and a T-cell line derived from an ADA-deficient severe combined immunodeficiency (SCID) patient demonstrate that the SL3-3 chLTR is the most active in these lymphoid cell lines. The greatest difference in the comparison of the different chLTR vectors was obsd. in primary human umbilical vein endothelial cells, where the MoMLV vector produced up to 100 times more CAT activity than the SL3-3 vector. These data suggest that the use of specific promoter/enhancer elements may lead to higher levels of gene expression following retroviral-mediated gene transfer into specific cell types and these observations may be useful in the design of human gene therapy expts.

L14 ANSWER 4 OF 21 HCA COPYRIGHT 1995 ACS
AN 122:96039 HCA
TI Development of live tumor vaccines using **retroviral vectors** for transfer of suicide genes and cytokines
AU **Mullen, Craig A.**
CS Diagnosis and Centers, National Cancer Institute, Bethesda, MD, USA
SO Contrib. Oncol. (1994), 46(CYTOKINES IN CANCER THERAPY), 260-8

CODEN: COONEV; ISSN: 0250-3220
DT Journal
LA English
AB Recent work with suicide genes and cytokines indicates the following:. Animals can be treated with large doses of the prodrug 5-fluorocytosine (5-FC) without serious toxicity. Live cytosine deaminase (CD)+ tumors can be eliminated in vivo by 5-FC treatment, suggesting that live attenuated strains of tumor can be produced. Animals rejecting CD+ tumors after 5-FC treatment develop immunity to wild-type tumor. Tumors modified to secrete IL-6 are rejected or grow more slowly than wild-type tumor. Animals rejecting IL-6-secreting tumor also develop immunity to wild-type tumor. Double transduction of tumor with CD and IL-6 genes allows delivery of much larger doses of cytokine-secreting tumor to the host. These results suggest that live attenuated tumor vaccines can be produced with gene transfer techniques and that work to find optimal combinations of genes and immunization schedules is justified.

L14 ANSWER 5 OF 21 HCA COPYRIGHT 1995 ACS
AN 122:45771 HCA
TI Inhibition of HIV-1 in human T-lymphocytes by retrovirally transduced anti-tat and rev hammerhead ribozymes
AU Zhou, Chen; Bahner, Ingrid C.; Larson, Garry P.; Zaia, John A.; Rossi, John J.; **Kohn, Donald B.**
CS Division of Research Immunology and Bone Marrow Transplantation, Childrens Hospital Los Angeles, Departments of Pediatrics and Microbiology, University of Southern California School of Medicine, Los Angeles, CA, 90027, USA
SO Gene (1994), 149(1), 33-9
CODEN: GENED6; ISSN: 0378-1119
DT Journal
LA English
AB Gene therapy for AIDS requires the identification of genes which effectively inhibit HIV-1 replication coupled to an efficient vector system for gene delivery and expression. Hammerhead ribozymes are RNA mols. capable of catalytic cleavage of complementary RNA mols. Ribozymes targeted against two portions of the HIV-1 genome were designed to cleave HIV RNA in the tat gene (TAT) or in a common exon for tat and rev (TR). The ribozymes were cloned into the LN (LTR-neomycin) retroviral vector plasmids and expressed as part of viral LTR-driven transcripts. The vectors were packaged as amphitropic virions and used to transduce human T-lymphocytes. Expression of the vector transcripts contg. the ribozyme sequences was readily detected by Northern blot anal. of the transduced T cells. The T-lymphocytes expressing the anti-HIV-1 ribozymes showed resistance to HIV-1 replication. In contrast, cells expressing mutant ribozymes, contg. substitutions of a key nucleotide in the catalytic domain which cripples the cleavage activity of the ribozymes, supported replication of HIV-1, demonstrating that the functional ribozymes were cleaving the target RNAs. These studies demonstrate that retrovirally transduced ribozymes included in long, multifunctional transcripts, can inhibit HIV replication in human T-lymphocytes. The ribozyme and expression strategies described here should be useful for the gene therapy of AIDS by conferring resistance to HIV-1 replication on cells derived from transduced

hematopoietic stem cells.

L14 ANSWER 6 OF 21 HCA COPYRIGHT 1995 ACS
AN 120:316888 HCA
TI Lack of expression from a **retroviral vector** after transduction of murine hematopoietic stem cells is associated with methylation in vivo
AU Challita, Pia Maria; **Kohn, Donald B.**
CS Sch. Med., Univ. Southern California, Los Angeles, CA, 90027, USA
SO Proc. Natl. Acad. Sci. U. S. A. (1994), 91(7), 2567-71
CODEN: PNASA6; ISSN: 0027-8424
DT Journal
LA English
AB The authors describe studies of gene transfer and expression of the human glucocerebrosidase cDNA by a Moloney murine leukemia virus (MoMuLV)-based retroviral vector in a murine gene transfer/bone marrow transplant (BMT) model. Pluripotent hematopoietic stem cells (HSCs) were assayed as the colony-forming units, spleen (CFU-S) generated after serial transplantation. Transcriptional expression from the MoMuLV long-terminal repeat (LTR) was detected at a high level in the primary (1.degree.) CFU-S and tissues of reconstituted BMT recipients. However, the authors obsd. transcriptional inactivity of the proviral MoMuLV-LTR in >90% of the secondary (2.degree.) CFU-S and in 100% of the tertiary (3.degree.) CFU-S examd. The authors have compared the methylation status of the provirus in the 1.degree. CFU-S, which show strong vector expression, to that of the transcriptionally inactive provirus in the 2.degree. and 3.degree. CFU-S by Southern blot anal. using the methylation-sensitive restriction enzyme Sma I. The studies demonstrated a 3- to 4-fold increase in methylation of the SmaI site in the proviral LTR of 2.degree. and 3.degree. CFU-S compared to the transcriptionally active 1.degree. CFU-S. These observations may have important implications for future clin. applications of retroviral-mediated gene transfer into HSCs, where persistent gene expression would be needed for an enduring therapeutic effect.

L14 ANSWER 7 OF 21 HCA COPYRIGHT 1995 ACS
AN 120:126753 HCA
TI Growth factors increase amphotropic retrovirus binding to human **CD34**+ bone marrow progenitor cells
AU Crooks, Gay M.; **Kohn, Donald B.**
CS Div. Res. Immunol., Child. Hosp. Los Angeles, Los Angeles, CA, USA
SO Blood (1993), 82(11), 3290-7
CODEN: BLOOAW; ISSN: 0006-4971
DT Journal
LA English
AB Gene transfer into human cells using murine amphotropic retroviral vectors is the basic technique used in most current gene therapy studies. The identity of the cell surface receptor for the amphotropic envelope remains unknown and thus its importance in gene transfer is poorly understood. The authors have measured specific retrovirus binding to cells to study amphotropic virus receptor regulation in human CD34+ CD38- human hematopoietic cells. The rat monoclonal antibody 83A25 recognizes an epitope common to the envelope glycoprotein of all classes of Mononey murine leukemia

virus. Indirect fluorescent labeling of 83A25 allows flow cytometric anal. of specific virus-cell interactions and is an indirect measure of specific receptors. Using this assay, amphotropic virus binding to fresh CD34+ cells were minimal. However, when CD34+ cells were cultured with or without growth factors for 4 days, specific binding of amphotropic retrovirus was readily shown. Inclusion of interleukin-3 (IL-3), IL-6, and Steel factor in cultures increased the fluorescence assocd. with amphotropic virus binding by 2-4 fold (mean fold increase 2.7 .+-. 0.84). Virus binding to CD34+ CD38- cells was shown only in those cells cultured in IL-3, IL-6, and Steel factor. These results suggest that certain cytokines may cause an increase in the no. and/or affinity of amphotropic receptors on primitive human hematopoietic cells. Upregulation of viral receptor expression may be one of the mechanisms by which cytokines enhance gene transfer into primitive BM cells.

L14 ANSWER 8 OF 21 HCA COPYRIGHT 1995 ACS
AN 120:23215 HCA
TI T lymphocyte ontogeny in **adenosine deaminase** -deficient **severe combined immune deficiency** after treatment with polyethylene glycol-modified **adenosine deaminase**
AU Weinberg, Kenneth; Hershfield, Michael S.; Bastian, John; **Kohn, Donald**; Sender, Leonard; Parkman, Robertson; Lenarsky, Carl
CS Sch. Med., Univ. South. California, Los Angeles, CA, 90027, USA
SO J. Clin. Invest. (1993), 92(2), 596-602
CODEN: JCINAO; ISSN: 0021-9738
DT Journal
LA English
AB Adenosine deaminase (ADA) deficiency causes severe combined immune deficiency (SCID) by interfering with the metab. of deoxyadenosine, which is toxic to T lymphocytes at all stages of differentiation. Enzyme replacement with polyethylene glycol-modified ADA (PEG-ADA) has been previously shown to correct deoxyadenosine metab. and improve mitogen-induced T lymphocyte proliferation. The authors studied the biochem. and immunol. effects of PEG-ADA in two infants with ADA-deficient SCID. While in a catabolic state, higher doses of PEG-ADA than previously described were required to normalize deoxyadenosine metab. After biochem. improvement, the patients recovered immune function in a pattern similar to that obsd. in normal thymic ontogeny and in patients with immunol. reconstitution after bone marrow transplantation. Immune reconstitution was marked by the sequential appearance in the peripheral blood of phenotypic T lymphocytes corresponding to successive stages of thymic differentiation. Functional reconstitution was marked by the successive appearance of mitogen responses dependent on exogenous in vitro IL-2, mitogen responses not requiring exogenous IL-2, antigen-specific responses dependent on exogenous IL-2, and finally, antigen-specific responses not requiring exogenous IL-2. Natural killer function was tested in one patient and normalized with PEG-ADA therapy. Optimal PEG-ADA therapy appears to normalize thymic differentiation in ADA-deficient SCID, resulting in normal antigen-specific immune function.

L14 ANSWER 9 OF 21 HCA COPYRIGHT 1995 ACS
AN 119:87556 HCA
TI Comparison of trans-dominant inhibitory mutant human immunodeficiency virus type 1 genes expressed by **retroviral vectors** in human T lymphocytes
AU Bahner, Ingrid; Zhou, Chen; Yu, Xiao-Jin; Hao, Qian-Lin; Guatelli, John C.; **Kohn, Donald B.**
CS Div. Res. Immunol./Bone Marrow Transplant., Child. Hosp., Los Angeles, CA, 90027, USA
SO J. Virol. (1993), 67(6), 3199-207
CODEN: JOVIAM; ISSN: 0022-538X
DT Journal
LA English
AB Trans-dominant inhibitory mutant versions of the human immunodeficiency virus type 1 (HIV-1) regulatory genes tat and rev have previously been described. The authors have constructed a series of retroviral vectors to transduce these genes and compare their inhibitory activities. The inhibitory activities were measured with transient transfection assays by using a reporter which expresses an HIV-1 gag-Escherichia coli lacZ fusion protein with strict dependence on coexpression of both tat and rev. Addnl., the vectors were packaged as amphotropic virions and used to stably transduce human CEM T lymphocytes. The transduced CEM cells were challenged with HIV-1, and the effects of the mutant HIV-1 genes were detd. by measuring the levels of HIV-1 p24gag produced. A tat gene substituted at amino acid 41 (tatk41a) retained partial trans-activating activity and lacked inhibitory activity. A tat gene with a premature stop codon at amino acid 54 (tat54ter) showed moderate trans-dominant inhibition of the reporter plasmid but failed to significantly inhibit HIV-1 replication. The M10 rev mutant, with a 2-amino-acid substitution, showed strong trans-dominant inhibitory activity both in the reporter plasmid and in the HIV-1 infection assay. The greatest inhibition of HIV-1 growth was seen when M10 was expressed under the transcriptional control of a human cytomegalovirus promoter; slightly less inhibition was achieved when expression of M10 was controlled by the Moloney murine leukemia virus long terminal repeat, and minimal inhibition was seen when the HIV-1 long terminal repeat controlled the M10 gene. These results demonstrate the potential utility of retroviral vectors expressing trans-dominant inhibitory mutant HIV-1 genes for gene therapy approaches to AIDS.

L14 ANSWER 10 OF 21 HCA COPYRIGHT 1995 ACS
AN 118:52210 HCA
TI Retroviral-mediated transfer of the human glucocerebrosidase gene into cultured Gaucher bone marrow. [Erratum to document cited in CA117(15):143214t]
AU Nolta, Jan A.; Yu, Xiao Jin; Bahner, Ingrid; **Kohn, Donald B.**
CS Dep. Pediatr., Child. Hosp. Los Angeles, Los Angeles, CA, 90027, USA
SO J. Clin. Invest. (1992), 90(4), 1635
CODEN: JCINAO; ISSN: 0021-9738
DT Journal
LA English
AB An error in Figure 2 has been cor. The error was not reflected in the abstr. or the index entries.

L14 ANSWER 11 OF 21 HCA COPYRIGHT 1995 ACS
AN 117:143214 HCA
TI Retroviral-mediated transfer of the human glucocerebrosidase gene into cultured Gaucher bone marrow
AU Nolta, Jan A.; Yu, Xiao Jin; Bahner, Ingrid; **Kohn, Donald B.**
CS Dep. Pediatr., Child. Hosp. Los Angeles, Los Angeles, CA, 90027, USA
SO J. Clin. Invest. (1992), 90(2), 342-8
CODEN: JCINAO; ISSN: 0021-9738
DT Journal
LA English
AB Gaucher disease, a lysosomal glycolipid storage disorder, results from the genetic deficiency of an acidic glucosidase, glucocerebrosidase (GC). The beneficial effects of allogeneic bone marrow transplantation (BMT) for Gaucher disease suggest that GC gene transduction and the transplantation of autologous hematopoietic stem cells (gene therapy) may similarly alleviate symptoms. The authors constructed a retroviral vector, L-GC, produced by a clone of the amphotropic packaging cell line PA317, which transduces the normal human GC cDNA with high efficiency. Whole-marrow mononuclear cells and CD34-enriched cells from a 4-yr-old female with type 3 Gaucher disease were transduced by the L-GC vector and studied in long-term bone marrow culture (LTBMC). Prestimulation of marrow with IL-3 and IL-6, followed by co-cultivation with vector-producing fibroblasts, produced gene transfer into 40-45% of the hematopoietic progenitor cells. The levels of GC expression in progeny cells (primarily mature myelomonocytic) produced by the LTBMC were quant. analyzed by Northern blot, Western blot, and glucocerebrosidase enzyme assay. Normal levels of GC RNA, immunoreactive protein, and enzymic activity were detected throughout the duration of culture. These studies demonstrate that retroviral vectors can efficiently transfer the GC gene into long-lived hematopoietic progenitor cells from the bone marrow of patients with Gaucher disease and express physiol. relevant levels of GC enzyme activity.

L14 ANSWER 12 OF 21 HCA COPYRIGHT 1995 ACS
AN 112:133770 HCA
TI Expression of human glucocerebrosidase in murine long-term bone marrow cultures after **retroviral vector**-mediated transfer
AU Nolta, Jan A.; Sender, Leonard S.; Barranger, John A.; **Kohn, Donald B.**
CS Div. Res. Immunol., Child. Hosp. Los Angeles, Los Angeles, CA, USA
SO Blood (1990), 75(3), 787-97
CODEN: BLOOAW; ISSN: 0006-4971
DT Journal
LA English
AB A retroviral vector (N2-SV-GC) was constructed by inserting a normal human glucocerebrosidase (GC) cDNA under control of the SV40 early region promoter into the Moloney murine leukemia virus-derived N2 vector. N2-SV-GC produced human GC in murine 3T3 fibroblasts at levels in the range of the endogenous murine GC as detd. by enzymic assay and Western blot anal. The N2-SV-GC retroviral vector was used for studies of gene transduction of murine hematopoietic

progenitor cells (HPC). Infection of bone marrow cultured for 2 to 10 days in medium contg. hematopoietic growth factors was significantly more efficient than infection of freshly isolated marrow cells (24% to 32% G418-resistant CFU-GM 15%, resp.). The marrow infected by N2-SV-GC was maintained in long-term bone marrow culture (LTBMCe and had a stable level of G418-resistant HPC over 2 mo of serial assays. The human GC gene of the vector was persistently expressed in the nonadherent cell fraction of the murine LTBMC as detd. by Northern blotting, Western blotting, and immunohistochem. staining using a monoclonal antibody specific for human GC. N2-SV-GC also expressed the human GC gene in day 12 CFU-S. LTBMC represents a novel system for retroviral vector-mediated gene transduction of HPC and may accurately predict the activities of vectors in vivo.

L14 ANSWER 13 OF 21 HCA COPYRIGHT 1995 ACS photocopy (AKM)
AN 111:210043 HCA
TI Transfer and expression of the human **adenosine **deaminase*** (ADA) gene in ADA-deficient human T lymphocytes with **retroviral vectors**
AU **Kohn, Donald B.**; Kantoff, Philip; Zwiebel, James; Gilboa, Eli; Anderson, W. French; Blaese, R. Michael
CS Metab. Branch, NCI, Bethesda, MD, 20892, USA
SO UCLA Symp. Mol. Cell. Biol., New Ser. (1989), 87(Gene Transfer Gene Ther.), 365-74
CODEN: USMBD6; ISSN: 0735-9543
DT Journal
LA English
AB Transfer of the human ADA gene into an HTLV-1 transformed, ADA-deficient human T lymphocyte line (TJF-2) by a retroviral vector (SAX) was previously shown to produce normal levels of ADA activity. This report describes SAX infection of non-transformed T lymphocytes from ADA-deficient patients. This leads to increased levels of ADA activity in these primary T cells, similar to those produced in the transformed T line. To quantitate the rate of ADA gene transfer and expression by SAX, TJF-2 cells which were infected by SAX were cloned by limiting diln. Six out of 27 (22%) of the clones had acquired and were expressing the SAX vector. One to 3 copies of SAX/cell produced ADA activity in the range found in normal thymocytes and T lymphocytes. Similar vectors with other promoters were also highly active. Thus, these vectors are capable of efficient transfer and expression of the human ADA gene in ADA-deficient, human T cells.

L14 ANSWER 14 OF 21 HCA COPYRIGHT 1995 ACS
AN 111:188732 HCA
TI Expression of the human glucocerebrosidase gene by **retrovirus vectors**
AU **Kohn, Donald B.**; Nolta, Jan A.; Hong, Chang Mu; Barranger, John A.
CS Div. Res. Immunol. Bone Marrow Transplantat., Child. Hosp. Los Angeles, Los Angeles, CA, 90027, USA
SO UCLA Symp. Mol. Cell. Biol., New Ser. (1989), 87(Gene Transfer Gene Ther.), 397-408
CODEN: USMBD6; ISSN: 0735-9543

DT Journal
LA English
AB Gaucher disease, caused by glucocerebrosidase deficiency, may be a candidate for early trials of human gene therapy. A series of retrovirus vectors, which contain either a normal human glucocerebrosidase cDNA or a minigene with a 5' genomic glucocerebrosidase gene fragment fused to the 3' portion of the cDNA was constructed. These genes were under transcriptional control of either heterologous flanking region. Transfection or infection of the vectors into murine fibroblasts results in expression of glucocerebrosidase activity to levels equal to that of normal murine or human fibroblasts. The rank order of promoter activity is: glucocerebrosidase > SV40 > thymidine kinase. The conferred glucocerebrosidase activity is immunoprecipitable by a monoclonal antibody specific for the human enzyme. Western blots show the expressed protein is of the expected size range (59-66 kd). Southern blotting reveals that cells which express activity in the normal range contain a single intact copy of the vector. Retrovirus vectors are capable of high efficiency transduction of the human glucocerebrosidase gene and may be useful for clin. gene therapy.

L14 ANSWER 15 OF 21 HCA COPYRIGHT 1995 ACS
AN 111:110248 HCA
TI Retroviral-mediated gene transfer into hemopoietic cells
AU Eglitis, Martin A.; Kantoff, Philip W.; **Kohn, Donald B.**; Karson, Evelyn; **Moen, Robert C.**; Lothrop, Clinton D., Jr.; Blaese, R. Michael; Anderson, W. French
CS Lab. Mol. Hematol., NIH, Bethesda, MD, USA
SO Adv. Exp. Med. Biol. (1988), 241(Mol. Biol. Hemopoiesis), 19-27
CODEN: AEMBAP; ISSN: 0065-2598
DT Journal
LA English
AB Retroviral vectors have provided a means for the introduction of functioning exogenous genes into the hematopoietic system of whole animals. Although these vectors are quite efficient in the mouse model, when applied to non-murine in vivo systems, the efficiency of gene transfer has diminished to impractical levels. Since in vivo analyses are expensive and time consuming, in vitro models have been developed to speed the evaluation of alternative protocols. Using in vitro colony assays, 3 approaches were evaluated for their ability to improve the infectivity of hematopoietic progenitor cells with retorviral vectors. Exogenously applied hematopoietic growth factors increased the proportion of hematopoietic colones in vitro up to an av. of 5-fold. when alternative sources of progenitors, such as fetal cord blood, were used, improvements in infection efficiency were also obtained. Finally, evidence was acquired suggesting that xenotropic packaging of vectors also improved infection efficiency.

photocopy - milne

L14 ANSWER 16 OF 21 HCA COPYRIGHT 1995 ACS
AN 111:22025 HCA
TI Establishment and characterization of **adenosine deaminase**-deficient human T cell lines
AU **Kohn, Donald B.**; Mitsuya, Hiroaki; Ballow, Mark; Selegue, Jane E.; Barankiewicz, Jerzy; Cohen, Amos; Gelfand, Erwin; Anderson,

10% has been found 153 days after transplantation. Human bone marrow has also been treated with the N2 vector, resulting in 1-2% G418-resistant progenitors.

L14 ANSWER 18 OF 21 HCA COPYRIGHT 1995 ACS
AN 107:212900 HCA
TI Retroviral-mediated gene transfer into mammalian cells
AU **Kohn, Donald B.**; Kantoff, Philip W.; Eglitis, Martin A.; McLachlin, Jeanne R.; **Moen, Robert C.**; Karson, Evelyn; Zwiebel, James A.; Nienhuis, Arthur; Karlsson, Stefan; et al.
CS Metab. Branch, Natl. Cancer Inst., Bethesda, MD, 20892, USA
SO Blood Cells (1987), 13(1-2), 285-98
CODEN: BLCEDD; ISSN: 0340-4684
DT Journal
LA English
AB Retroviruses may be used as genetic vectors to transfer genes into mammalian cells with high efficiency. The N2 vector will transfer a functional bacterial gene for neomycin resistance (NeoR) into more than 80% of mouse spleen foci. A deriv. of the N2 vector was constructed to study transfer and expression of the human gene for adenosine deaminase (ADA) in mammalian lymphoid and hematopoietic stem cells. This vector, termed SAX, contains the human ADA cDNA with an SV40 promoter in addn. to the NeoR gene. The SAX vector was found to efficiently transfer and express the ADA gene in an ADA-deficient human T-cell line. Gene transfer by SAX using an autologous nonhuman primate bone marrow transplant model resulted in expression of the human ADA gene in peripheral blood cells of treated animals. Human bone marrow treated with SAX produced 1-2% of colonies in vitro that were expressing the vector genes. Transfer of genes into circulating hematopoietic stem cells of fetal sheep in utero was most efficient; vector gene expression was evident in 20-40% of hematopoietic colonies. Therefore, retroviral vectors are capable of transferring functional genes into a wide variety of mammalian lymphoid and hematopoietic cells. Such vectors may be useful for clin. trials of gene therapy, i.e., the correction of genetic diseases by insertion of a normal gene into a patient's defective cells.

L14 ANSWER 19 OF 21 HCA COPYRIGHT 1995 ACS
AN 107:91223 HCA
TI Expression of human **adenosine deaminase** in nonhuman primates after retrovirus-mediated gene transfer
AU Kantoff, Philip W.; Gillio, Alfred P.; McLachlin, Jeanne R.; Bordignon, Claudio; Eglitis, Martin A.; Kernan, Nancy A.; **Moen, Robert C.**; **Kohn, Donald B.**; Yu, Sheau Fung; et al.
CS Lab. Mol. Hematol., Natl. Heart, Lung, Blood Inst., Bethesda, MD, 20892, USA
SO J. Exp. Med. (1987), 166(1), 219-34
CODEN: JEMEAV; ISSN: 0022-1007
DT Journal
LA English
AB Primate bone marrow cells were infected with a retroviral vector carrying the genes for human adenosine deaminase (h-ADA) and bacterial neomycin resistance (neor). The infected cells were infused back into the lethally irradiated donor animals. Several

monkeys fully reconstituted and expressed the h-ADA and neor genes at low levels in their recirculating hematopoietic cells for short periods of time.

L14 ANSWER 20 OF 21 HCA COPYRIGHT 1995 ACS
AN 107:34292 HCA
TI Retroviral-mediated gene transfer into hematopoietic cells
AU Kantoff, Philip W.; Gillio, Al; McLachlin, Jeanne R.; Flake, Alan W.; Eglitis, Martin A.; **Moen, Robert**; Karlsson, Stefan; Kohn, Don B.; Karson, Evelyn; et al.
CS Lab. Mol. Hematol., Natl. Heart, Lung, Blood Inst., Bethesda, MD, 20892, USA
SO Trans. Assoc. Am. Physicians (1986), 99, 92-102
CODEN: TAAPAI; ISSN: 0066-9458
DT Journal
LA English
AB Protocols for gene transfer using bone marrow transplantation were developed in 2 large animal models, nonhumanprimates (rhesus monkey and cynomolgus monkey) and fetus of sheep. Retroviral vectors, N2 or SAX, carrying the transposon Tn5 neomycin resistance gene or the neomycin resistance gene and the human adenosine deaminase cDNA, resp., were transferred to isolated bone marrow cells by either co-cultivation with virus-producing fibroblasts or suspension in virus-contg. supernatant from virus-producing cells. After transfection, the bone marrow cells were washed and infused back into the donor animal. Reconstitution represented the full recovery of all images. Monkeys whose bone marrow cells were subjected to co-cultivation failed to fully reconstitute. However, after the supernatant infection protocol, the bone marrow cells showed full reconstitution and low but clearly detectable and reproducible levels of neomycin resistance and human adenosine deaminase gene activity. Circulating hematopoietic cells from a 100-day-old fetal lamb were removed in utero by exchange transfusion, infected with the N2 vector by the supernatant infection protocol, and infused back into the lamb. After the birth of the lamb, G418 resistant bone marrow cells were obtained from the infected animal but not from age-matched control lambs. When human bone marrow cells were transfected with the N2 vector by either the co-cultivation or the supernatant protocol, the levels of functional gene transfer was 1-2%. The potential utility of these methods in establishing a human gene therapy protocol is discussed.

L14 ANSWER 21 OF 21 HCA COPYRIGHT 195 ACS
AN 105:166144 HCA
TI Correction of **adenosine deaminase** deficiency in cultured human T and B cells by retrovirus-mediated gene transfer
AU Kantoff, Philip W.; **Kohn, Donald B.**; Mitsuya, Hiroaki; Armentano, Donna; Sieberg, Miri; Zwiebel, James A.; Eglitis, Martin A.; McLachlin, Jeanne R.; Wiginton, Dan A.; et al.
CS Lab. Mol. Hematol., Natl. Heart, Lung, Blood Inst., Bethesda, MD, 20892, USA
SO Proc. Natl. Acad. Sci. U. S. A. (1986), 83(17), 6563-7
CODEN: PNASA6; ISSN: 0027-8424
DT Journal
LA English

AB A retroviral vector called SAX, contg. the cloned human cDNA for adenosine deaminase (ADA) [9026-93-1], was constructed and used to introduce the ADA gene into cultured T- and B-lymphocyte lines derived from patients with ADA deficiency. DNA anal. showed that the SAX vector was inserted intact into the T and B cells at .apprx.1 copy per cell. The treated cells produced the characteristic isoenzymes of human ADA at a level similar to normal T and B lymphocytes. It is known that ADA-deficient lymphocytes are unusually sensitive to high levels of 2'-deoxyadenosine, and this is the mechanism though to underlie the selective lymphocytotoxicity assocd. with ADA deficiency in vivo. Expression of the introduced ADA gene was sufficient to reverse the hypersensitivity of these genetically deficient lymphocytes to 2'-deoxyadenosine toxicity. These results support the suggestion that retroviral vector gene-delivery systems show promise for application to human gene therapy.

L16 ANSWER 1 OF 2 HCA COPYRIGHT 1995 ACS
AN 120:321192 HCA
TI Gene transfer into nonhuman primate **CD34**+CD11b-bone marrow progenitor cells capable of repopulating lymphoid and myeloid lineages
AU Van Beusechem, Victor W.; Bart-Baumeister, Julia A. K.; Bakx, Trudy A.; Kaptein, Leonie C. M.; Levinsky, Roland J.; Valerio, Dinko
CS TNO Med. Biol. Lab., Dep. Gene Ther., Rijswijk, Neth.
SO Hum. Gene Ther. (1994), 5(3), 295-305
CODEN: HGTHE3; ISSN: 1043-0342
DT Journal
LA English
AB The authors investigated whether rhesus monkey CD34+CD11b- hematopoietic stem cells can be transduced with recombinant retroviruses carrying the human adenosine deaminase (hADA) gene by co-cultivation with a virus-producing cell line. Following autologous transplantation, polymerase chain reaction (PCR) anal. on peripheral blood mononuclear cells and granulocytes showed that the hADA-retrovirus was present in approx. 0.1% of the cells for at least 400 days post transplantation in 2 monkeys. Bone marrow that was harvested 16 mo after transplantation carried ADA-overexpressing myeloid progenitor cells capable of in vitro colony formation. In addn., hADA activity could be demonstrated in T lymphocytes that were harvested 9 mo post transplantation. Thus, in vitro transduction of CD34+CD11b- cells led to long-term repopulation of the hematopoietic system with transduced cells of lymphoid and myeloid lineages expressing the hADA gene. To investigate whether infusion of virus-producing cells into a rhesus monkey undergoing autologous bone marrow transplantation could lead to in vivo transfer of the recombinant retrovirus, 1 monkey was infused with CD34+CD11b- bone marrow cells (BMC) and a large quantity of virus-producing cells. Few provirus-carrying cells could temporarily be detected in this animal. This shows that in vivo gene transfer into a regenerating hemopoietic system can occur, albeit at very low efficiency.
CC 15-7 (Immunochemistry)

IT Transplant and Transplantation
(bone marrow, gene transfer into nonhuman primate **CD34** -pos. progenitor cells in cell repopulation after)
IT Transformation, genetic
(of genes, into nonhuman primate **CD34**-pos. progenitor cells, for cell repopulation after bone marrow transplant)
IT Hematopoietic precursor cell
(lymphoid, gene transfer into nonhuman primate **CD34** -pos., cell repopulation after bone marrow transplant of)
IT Hematopoietic precursor cell
(myeloid, gene transfer into nonhuman primate **CD34** -pos., cell repopulation after bone marrow transplant of)
IT **Virus**, animal
(**retro-**, **adenosine deaminase** gene transfer mediated by, into nonhuman primate **CD34**-pos. progenitor cells, for cell repopulation after bone marrow transplant)
IT Bone marrow
(transplant, gene transfer into nonhuman primate **CD34** -pos. progenitor cells in cell repopulation after)
IT 9026-93-1, **Adenosine deaminase**
(gene for human, transfer of, **retrovirus**-mediated, into nonhuman primate **CD34**-pos. progenitor cells, for cell repopulation after bone marrow transplant)

L16 ANSWER 2 OF 2 HCA COPYRIGHT 1995 ACS
AN 120:69038 HCA
TI Long-term in vivo expression of a murine **adenosine deaminase** gene in rhesus monkey hematopoietic cells of multiple lineages after **retroviral** mediated gene transfer into **CD34**+ bone marrow cells
AU Bodine, David M.; Moritz, Tom; Donahue, Robert E.; Luskey, Barry D.; Kessler, Steven W.; Martin, David I. K.; Orkin, Stuart H.; Nienhuis, Arthur W.; Williams, David A.
CS Clin. Hematol. Branch, Natl. Heart, Lung, Blood Inst., Bethesda, MD, 20892, USA
SO Blood (1993), 82(7), 1975-80
CODEN: BLOOAW; ISSN: 0006-4971
DT Journal
LA English
AB Retroviral mediated gene transfer into stem cells has been proposed as therapy for many inherited hematopoietic diseases. Deficiency of the enzyme adenosine deaminase (ADA) results in depletion of T lymphocytes, causing severe combined immunodeficiency syndrome (SCIDS). In this report, the authors describe retroviral mediated gene transfer of a murine ADA cDNA into Rhesus monkey hematopoietic stem cells. Immunoselected CD34+ bone marrow cells were exposed to medium contg. the ADA retrovirus during culture on a stromal cell line engineered to express the transmembrane form of stem cell factor. After infusion of autologous, transduced cells into irradiated recipients, gene transfer was obsd. in all three monkeys. The ADA provirus was detected in 2% of circulating granulocytes and T cells from 100 days post-transplantation to longer than 1 yr and in B cells from 250 days post-transplantation and beyond. Mouse ADA activity was detected in peripheral blood cells at approx. 3% the

activity of monkey ADA. Thus, the authors have shown gene transfer into repopulating cells that contribute to all hematopoietic lineages with persistent gene expression. These data provide support for the use of stem cell targeted gene transfer for therapy of ADA deficiency.

CC 1-7 (Pharmacology)
Section cross-reference(s): 3

ST ADA deficiency gene therapy **retrovirus** vector; hematopoietic stem cell ADA gene transfer; **adenosine deaminase** deficiency gene therapy

IT Hematopoietic precursor cell
(**adenosine deaminase** gene of rats transfer into Rhesus monkey, **retro virus**-mediated, gene therapy of severe combined immunodeficiency syndrome in relation to)

IT Gene, animal
(for **adenosine deaminase, retro virus**-mediated transfer of murine, to Rhesus monkey hematopoietic cells, gene therapy of severe combined immunodeficiency syndrome in relation to)

IT Transduction, genetic
(of **adenosine deaminase** gene of rats into Rhesus monkey hematopoietic cells)

IT Genetic vectors
(**retroviral**, for rat **adenosine deaminase** gene transfer into rhesus monkey **CD34** + bone marrow cells, enzyme gene expression in hematopoietic cells in relation to)

IT Lymphocyte
(T-cell, **adenosine deaminase** of, after murine gene transfer into Rhesus monkey hematopoietic cells)

IT Therapeutics
(geno-, for **adenosine deaminase** deficiency and severe combined immunodeficiency syndrome, **retro virus**-mediated of murine **adenosine deaminase** gene transfer into Rhesus monkey hematopoietic cells in)

IT Immunodeficiency
(severe combined, gene therapy for, **adenosine deaminase** gene transfer into hematopoietic cells in relation to)

IT 9026-93-1, **Adenosine deaminase**
(gene for, **retro virus**-mediated transfer of murine to Rhesus monkey hematopoietic cells, severe combined immunodeficiency syndrome gene therapy in relation to)

=> fil biosis
FILE 'BIOSIS' ENTERED AT 08:50:01 ON 10 JUL 95

FILE COVERS 1969 TO DATE.
CAS REGISTRY NUMBERS AND CHEMICAL NAMES (CNs) PRESENT FROM JANUARY 1969 TO DATE.

RECORDS LAST ADDED: 5 July 1995 (950705/ED)

CAS REGISTRY NUMBERS (R) LAST ADDED: 5 July 1995 (950705/UP)

As of December 31, 1993 the BIOSIS File will be updated weekly with information from both publications. SDIs will now be run weekly. For more information enter HELP UPDATE and HELP COST at an arrow prompt(=>).

=> d his l19-

(FILE 'BIOSIS' ENTERED AT 08:13:13 ON 10 JUL 95)

		E	KOHN D/AU
L19	224	S	E3-9
		E	BLAESE M/AU
L20	20	S	E3-4
		E	MULLEN C/AU
L21	43	S	E3-7
		E	MOEN R/AU
L22	82	S	E3-6
L23	359	S	L19 OR L20 OR L21 OR L22
L24	1983	S	(RETROVIR? OR RETRO (2W) VIR##) (3A) VECTOR#
L25	5021	S	ADENOSINE DEAMINASE
L26	1874	S	CD34
L27	88	S	L23 AND (L24 OR L25 OR L26)
L28	66	S	L23 AND L24
L29	8	S	L28 AND L25
L30	1	S	L29 AND L26
L31	196	S	SEVERE COMBINED IMMUNE DEFICIENCY
L32	3	S	L31 AND L23
L33	3	S	L24 AND L25 AND L26
L34	0	S	L33 AND L31
L35	2237	S	SEVERE COMBINED IMMUNODEFICIENCY
L36	16	S	L35 AND L23
L37	1	S	L35 AND L33
L38	6	S	L32 OR L33
L39	1	S	L37 OR L30
L40	5	S	L38 NOT L39

FILE 'HCA' ENTERED AT 08:48:45 ON 10 JUL 95

FILE 'BIOSIS' ENTERED AT 08:50:01 ON 10 JUL 95

=> d bib ab st l39;d bib ab st l40 1-5

L39 ANSWER 1 OF 1 BIOSIS COPYRIGHT 1995 BIOSIS
AN 94:32442 BIOSIS
DN 97045442
TI Strategies for gene therapy.
AU Blaese R M; **Mullen C A**; Ramsey W J
CS Cellular Immunol. Sect., Metabolism Branch, National Cancer Inst., National Inst. Health, Build. 10, Room 6B05, Bethesda, MD 20892, USA
SO Pathologie Biologie 41 (8). 1993. 672-676. ISSN: 0369-8114
LA English
AB The use of retroviral-mediated gene transfer to introduce a DNA label into T cells (TIL) being used in the immunotherapy of patients with malignant melanoma filially opened the door to the clinical

photocopy milne

application of gene therapy for a wide variety of inherited and acquired diseases. The gene therapy trial for ADA deficiency SCID has demonstrated that long-term stable expression of exogenous genes can be achieved in human T lymphocytes using **retroviral vectors** for ex vivo treatment and that significant immune reconstitution can be achieved in these patients following periodic infusions with ADA gene-corrected autologous T cells. Newer clinical applications include the insertion of genes into **CD34** enriched stem cell populations, the testing of autologous tumor vaccines employing cytokine acne-modified tumor cells and the direct transfer of the herpes thymidine kinase gene into brain tumors in situ in order to render those tumors sensitive to treatment with the ordinarily non-cytotoxic drug ganciclovir.

ST JOURNAL ARTICLE; HUMAN T-CELLS; RETROVIRAL-MEDIATED GENE TRANSFER; **SEVERE COMBINED IMMUNODEFICENCY**; **ADENOSINE DEAMINASE**; CANCER TREATMENT

L40 ANSWER 1 OF 5 BIOSIS COPYRIGHT 1995 BIOSIS
AN 95:236476 BIOSIS
DN 98250776
TI Correction of IL-4 receptor function in lymphoblastoid cell lines (LCL) from patients with X-linked **severe combined immune deficiency** (X-SCID) by retroviral mediated transfer of the gamma-C gene.
AU Uribe L; Taylor N; Smith S; Hong Y-H; Yu X-J; **Kohn D**; Weinberg K
CS Children's Hosp. Los Angeles, Los Angeles, CA, USA
SO 105th Annual Meeting of the American Pediatric Society and the 64th Annual Meeting of the Society for Pediatric Research, San Diego, California, USA, May 7-11, 1995. Pediatric Research 37 (4 PART 2). 1994. 11A. ISSN: 0031-3998
DT Conference
LA English
ST MEETING ABSTRACT; MEETING POSTER; INTERLEUKIN 4; GMMA-C GENE THERAPY

L40 ANSWER 2 OF 5 BIOSIS COPYRIGHT 1995 BIOSIS
AN 93:458859 BIOSIS
DN BA96:103759
TI T LYMPHOCYTE ONTOGENY IN ADENOSINE DEAMINASE-DEFICIENT **SEVERE COMBINED IMMUNE DEFICIENCY** AFTER TREATMENT WITH POLYETHYLENE GLYCOL-MODIFIED ADENOSINE DEAMINASE.
AU WEINBERG K; HERSHFIELD M S; BASTIAN J; **KOHN D**; SENDER L; PARKMAN R; LENARSKY C
CS DIV. RESEARCH IMMUNOL./BONE MARROW TRANSPLANTATION, CHILDRENS HOSPITAL LOS ANGELES, 4650 SUNSET BLVD. 62, LOS ANGELES, CA 90027, USA.
SO J CLIN INVEST 92 (2). 1993. 596-602. CODEN: JCINAO ISSN: 0021-9738
LA English
AB Adenosine deaminase (ADA) deficiency causes **severe combined immune deficiency** (SCID) by interfering with the metabolism of deoxyadenosine, which is toxic to T lymphocytes at all stages of differentiation. Enzyme replacement with polyethylene glycol-modified ADA (PEG-ADA) has been previously

population of pluripotent hematopoietic stem cells capable of self-renewal. We present evidence for the highly efficient gene transfer and sustained expression of human ADA in human primitive hematopoietic progenitors using retroviral supernatant with a supportive stromal layer. A stem cell-enriched (**CD34**+) fraction was also successfully transduced. Duchenne muscular dystrophy (DMD) is also a good model for somatic gene therapy. Two of the challenges presented by this model are the large size of the gene and the large number of target cells. Germline gene transfer and correction of the phenotype has been demonstrated in transgenic mdx mice using both a full-length and a truncated form of the dystrophin cDNA. We present here a deletion mutagenesis strategy to truncate the dystrophin cDNA such that it can be accommodated by **retroviral** and adenoviral **vectors** useful for somatic gene therapy.

ST HUMAN RETROVIRUS COMPLEMENTARY DNA **ADENOSINE DEAMINASE** PLURIPOTENT HEMATOPOIETIC STEM CELLS DUCHENNE MUSCULAR DYSTROPHY GENETIC ENGINEERING GENE THERAPY THERAPEUTIC METHOD

L40 ANSWER 5 OF 5 BIOSIS COPYRIGHT 1995 BIOSIS
AN 89:279851 BIOSIS
DN BR37:4848
TI **SEVERE COMBINED IMMUNE DEFICIENCY** SCID DUE TO DEFECTIVE INTERLEUKIN 2 RECEPTOR ALPHA IL2-R-A EXPRESSION.
AU WEINBERG K I; PARR T; ANNETT G M; COATES T; **KOHN D B**; LENARSKY C; PARKMAN R
CS DIV. RES. IMMUNOL. BONE MARROW TRANSPLANT., CHILD HOSP. LOS ANGELES, UNIV. SOUTH. CALIF., LOS ANGELES, CALIF., USA.
SO JOINT MEETING OF THE AMERICAN PEDIATRIC SOCIETY AND THE SOCIETY FOR PEDIATRIC RESEARCH, WASHINGTON, D.C., USA, MAY 1-4, 1989. PEDIATR RES 25 (4 PART 2). 1989. 170A. CODEN: PEREBL ISSN: 0031-3998
DT Conference
LA English
ST ABSTRACT HUMAN B LYMPHOCYTE T LYMPHOCYTE SIGNAL TRANSDUCTION

=> fil medlins
'MEDLINS' IS NOT A VALID FILE NAME
SESSION CONTINUES IN FILE 'BIOSIS'

=> fil medl
FILE 'MEDLINE' ENTERED AT 08:50:47 ON 10 JUL 95

FILE LAST UPDATED: 7 JUL 1995 (950707/UP). FILE COVERS 1966 TO DATE.
+QLF/CT SHOWS YOU THE ALLOWABLE QUALIFIERS OF A TERM.

MEDLINE, CANCERLIT AND PDQ ERRONEOUSLY ANNOTATED CERTAIN ARTICLES AUTHORED OR CO-AUTHORED BY DR. BERNARD FISHER WITH THE PHRASE "SCIENTIFIC MISCONDUCT-DATA TO BE REANALYZED." ALL SUCH ANNOTATIONS HAVE BEEN REMOVED OR ARE BEING REMOVED. WE APOLOGIZE FOR ANY PROBLEMS OR CONCERNS THIS MAY HAVE CAUSED. USERS SHOULD DISREGARD THOSE PRIOR ANNOTATIONS.

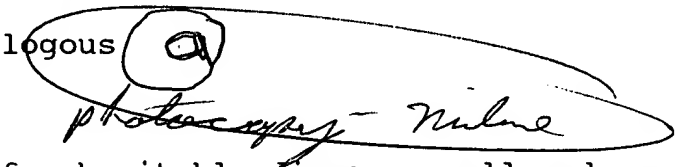
=> d his l41-

```
      (FILE 'HCA' ENTERED AT 08:48:45 ON 10 JUL 95)

      FILE 'BIOSIS' ENTERED AT 08:50:01 ON 10 JUL 95

      FILE 'MEDLINE' ENTERED AT 08:50:47 ON 10 JUL 95
L41         1409 S CD34
                 E ADENOSINE DEAMINASE/CT
L42         3191 S (ADENOSINE DEAMINASE+NT)/CT
                 E RETROVIRUS/CT
                 E RETROVIRIDAE/CT
                 E E3+ALL
                 E VECTORS/CT
                 E E3+ALL
                 E VECTORS/CT
                 E E8+ALL
                 E GENETIC VECTORS+NT/CT
                 E GENETIC VECTORS+ALL/CT
L43            7 S L41 AND L42
L44          375 S SEVERE COMBINED IMMUNODEFICIENCY+NT/CT
L45           67 S L44 AND L42 AND L42
L46           12 S L45 AND (VECTOR# OR RETROVIR?)
L47            2 S L46 AND L41

=> d bib ab ct 1-2

L47  ANSWER 1 OF 2  MEDLINE
AN   94002371     MEDLINE
TI   Treatment of severe combined immunodeficiency disease (SCID) due to
     adenosine deaminase deficiency with CD34+ selected
     autologous peripheral blood cells transduced with a human ADA gene.
     Amendment to clinical research project, Project 90-C-195, January
     10, 1992.
AU   Blaese R M; Culver K W; Chang L; Anderson W F; Mullen C; Nienhuis A;
     Carter C; Dunbar C; Leitman S; Berger M; et al
SO   Hum Gene Ther, (1993 Aug) 4 (4) 521-7.
     Journal code: A12. ISSN: 1043-0342.
CY   United States
DT   Journal; Article; (JOURNAL ARTICLE)
LA   English
FS   Priority Journals
EM   9401
AB   Significant increases in lymphocyte adenosine deaminase activity, T
     cell numbers and immune function have been achieved in the two
     children with SCID thus far treated with autologous T cells
     genetically-corrected by retroviral-mediated insertion of
     a normal ADA gene. Although the data obtained to date demonstrate
     that the use of ADA gene corrected peripheral T cells appears to be
     an effective treatment for ADA(-)SCID, it is theoretically
     preferable to try to develop a treatment for these children that
     will result in stem cell gene correction. The genetic correction of
     T cell progenitors with long-term immune reconstituting ability
     would be more desirable because repeated infusions of genetically
     altered cells should not be necessary and the generation of a more
     complete repertoire of T cell specificities might also be possible.
```

photocopy - milne

Furthermore, the present treatment protocol involves indefinite continuation of enzyme replacement treatment with PEG-ADA. The demonstration of ADA gene expression in the progeny of transduced stem cells may simplify the decision concerning cessation of this very costly enzyme treatment (approximately $250,000/yr./patient). Recent evidence suggests that a small fraction of bone marrow or peripheral blood mononuclear cells bearing the **CD34** antigen contains hematopoietic stem cells with both lymphoid and myeloid reconstituting ability. We propose in this amendment to supplement the infusion of human ADA gene-transduced autologous T cells in children with ADA(-)SCID with autologous peripheral blood **CD34**+ cells transduced with a second, readily distinguishable ADA **vector**.(ABSTRACT TRUNCATED AT 250 WORDS)

CT Check Tags: Human
Adenosine Deaminase: DF, deficiency
***Adenosine Deaminase: GE, genetics**
*Antigens, CD
Cells, Cultured
Clinical Protocols
*Gene Therapy
*Hematopoietic Stem Cell Transplantation
Hematopoietic Stem Cells: IM, immunology
Hematopoietic Stem Cells: ME, metabolism
*Hematopoietic Stem Cells: TR, transplantation
Severe Combined Immunodeficiency: EN, enzymology
Severe Combined Immunodeficiency: GE, genetics
***Severe Combined Immunodeficiency: TH, therapy**
T-Lymphocytes: IM, immunology
T-Lymphocytes: TR, transplantation
Transduction, Genetic
Transplantation, Autologous



L47 ANSWER 2 OF 2 MEDLINE
AN 93226692 MEDLINE
TI Gene transfer therapy for heritable disease: cell and expression targeting.
AU Mitani K; Clemens P R; Moseley A B; Caskey C T
CS Howard Hughes Medical Institute, Baylor College of Medicine, Houston, Texas 77030.
NC R01 DK42696 (NIDDK)
SO Philos Trans R Soc Lond B Biol Sci, (1993 Feb 27) 339 (1288) 217-24.
Ref: 70
Journal code: P5Z. ISSN: 0080-4622.
CY ENGLAND: United Kingdom
DT Journal; Article; (JOURNAL ARTICLE)
General Review; (REVIEW)
(REVIEW, TUTORIAL)
LA English
FS Priority Journals
EM 9307
AB Gene therapy is defined as the delivery of a functional gene for expression in somatic tissues with the intent to cure a disease. Different gene transfer strategies may be required to target different tissues. Adenosine deaminase (ADA) deficiency is a good

gene therapy model for targeting a rare population of pluripotent hematopoietic stem cells capable of self-renewal. We present evidence for the highly efficient gene transfer and sustained expression of human ADA in human primitive hematopoietic progenitors using **retroviral** supernatant with a supportive stromal layer. A stem cell-enriched (**CD34**+) fraction was also successfully transduced. Duchenne muscular dystrophy (DMD) is also a good model for somatic gene therapy. Two of the challenges presented by this model are the large size of the gene and the large number of target cells. Germline gene transfer and correction of the phenotype has been demonstrated in transgenic mdx mice using both a full-length and a truncated form of the dystrophin cDNA. We present here a deletion mutagenesis strategy to truncate the dystrophin cDNA such that it can be accommodated by **retroviral** and adenoviral **vectors** useful for somatic gene therapy.

CT Check Tags: Human; Support, Non-U.S. Gov't; Support, U.S. Gov't, P.H.S.

Adenosine Deaminase: DF, deficiency
Adenosine Deaminase: GE, genetics
Dystrophin: GE, genetics
*Gene Therapy: MT, methods
Genetic Vectors
*Hereditary Diseases: TH, therapy
Muscular Dystrophy: TH, therapy
Severe Combined Immunodeficiency: TH, therapy
Transfection: MT, methods